

# Acquisition of toxoplasma infection by children in a developing country

S. STAGNO<sup>1</sup> & E. THIERMANN<sup>2</sup>

*The prevalence of toxoplasma infection was studied in 850 healthy children (550 newborn infants and 300 children aged 6 months to 14 years) from a population of low socioeconomic status in Santiago, Chile. Antibodies were detected in 49 % of the serum samples in children at birth and in 0 % at 6 months, 2 % at 1 year, 10 % at 2 years, 22 % at 4 years, and 32 % at 6 years of age, and in 48 % of the children aged 7-14 years. In order to determine the rate of infection and associated epidemiological factors, 50 seronegative infants aged 6 months were followed up; 11 % and 14 % of them showed seroconversion after 1 and 2 years, respectively. The results suggest a significant association between infection and the ingestion of raw or undercooked meat. The role of cats and vectors such as flies and cockroaches in the transmission of infection could not be determined owing to the commonness of these two sources in relation to the small number of subjects studied.*

Infection with *Toxoplasma gondii* has been recognized in man and in many other vertebrates throughout the world by several authors (1, 3, 4, 9, 12-14, 16). These surveys revealed that the proportion of seropositive subjects (i.e., with antitoxoplasma antibodies) varied considerably with geographic area, eating habits, occupation, age, and probably with sanitary conditions and exposure to cats. The infection rates varied from less than 5 % in Navajo Indians, Australian aborigines, and Eskimos as reported by Feldman & Miller (4) to over 90 % in young women in Paris as demonstrated by Desmonts et al. (3).

With regard to the incidence of infection in childhood, the available information is based on a few longitudinal surveys. One study carried out in the USA by Warren & Dingle (22) on 40 families over a period of 10 years showed that only 5 % of the children developed antibodies to *Toxoplasma* during the course of the study. Paul (13) showed that the risk of infection in German children began at the age of 2 years and increased progressively to 6 years. An

association with eating undercooked meat was suggested by Desmonts (2) in Paris after studying the rate of appearance of antitoxoplasma antibodies in children in a tuberculosis hospital who were fed undercooked mutton. The rate of conversion from a negative to a positive antibody status was 4.8 % per month and increased to 9 % per month when two extra meals of undercooked meat were given every week.

In Chile, several investigators (12) have shown that the prevalence of *Toxoplasma* infections varied from 44 % in young adults living in the central valley to 85 % in island populations, including Easter Island. No data are available with respect to the incidence of infection in childhood. In the present study, the incidence of *Toxoplasma* infection and associated epidemiological factors were investigated in a group of normal children of low socioeconomic status.

## MATERIAL AND METHODS

### Study population

This consisted of 850 healthy children from birth up to 14 years of age from a low socioeconomic stratum living in different urban areas of Santiago, Chile. They were randomly selected among children attending a maternal and child health clinic and, at the time of inclusion in the study, were in good health and had no past history of malnutrition. The

<sup>1</sup> Instructor in Pediatrics and Parasitology, Faculty of Medicine, University of Chile, Santiago, Chile. Present address and address for reprints is UAB Medical Center, CDLD Room 609, University Station, Birmingham, Alabama 35294, USA.

<sup>2</sup> Assistant Professor of Parasitology, Faculty of Medicine, University of Chile, Santiago; Chief of the Laboratory of Toxoplasmosis, Department of Microbiology and Parasitology, University of Chile, Santiago, Chile.

study group comprised 550 newborn infants and 300 children divided into 6 groups of 50 aged 6 months, 1, 2, 4 and 6 years, and 7–14 (mean, 10) years.

To assess the rate of *Toxoplasma* infection, the children initially seen when 6 months old were also examined at 1½ years and again at 2½ years. At this time, a questionnaire was given to their mothers in order to determine their standard of living, the quality of their homes, the availability of potable water, the presence of domestic animals (cats, dogs, birds), the frequency of garbage collection, and the presence of vectors such as houseflies and cockroaches. The children's eating habits, with special regard to raw or undercooked meat, were also carefully examined, although no attempt was made to isolate *Toxoplasma* from meat.

At the first visit, a physical examination was carried out and a blood sample obtained from each child. The sera were then stored at  $-20^{\circ}\text{C}$  until examined, at which time the specimens were thawed and inactivated at  $56^{\circ}\text{C}$  for 30 min.

#### *Antitoxoplasma antibody tests*

The Sabin–Feldman dye test (SFT) was performed as described by Frenkel & Jacobs (6), and the indirect immunofluorescent antibody test (IFA) was carried out, as previously described by Stagno & Thiermann (18), with total anti-immunoglobulin from Baltimore Biological Laboratories. Contrast was enhanced and autofluorescence eliminated by counterstaining with 0.02% Evans' blue, which produced a red background. Four-fold dilutions were started at 1/4, but titres of 1/16 or more were considered to be positive in the present analysis. All patients with high antibody titres (1/512 or over) or who showed seroconversion (i.e., a change from a negative to a positive antibody status) were bled periodically and the physical examination including funduscopy was repeated. For statistical analysis the Chi-square test and the Pearson product-moment formula were used.

#### RESULTS

The agreement between the SFT and IFA results reached 98% (Table 1) and, in addition, there was close correlation between the actual titres obtained with the 2 tests ( $r = 0.71$ ). The present analysis is therefore based on the IFA results only.

#### *Prevalence of infection and antibody levels*

With regard to the prevalence of *Toxoplasma* infection, the rate of positive sera was 49% among

Table 1. Results of the indirect immunofluorescent test and the Sabin–Feldman dye test in 850 human sera

Indirect immuno-fluorescent test	Sabin–Feldman dye test		
	positive	negative	total
positive	325	10	335
negative	7	508	515
total	332	518	850

the newborn infants (Fig. 1). It dropped to 0% at 6 months and increased thereafter to reach 48% in the group of children aged 7–14 years. The distribution of antibody levels showed that titres of 1/512 or over were present in 10% of the newborn infants

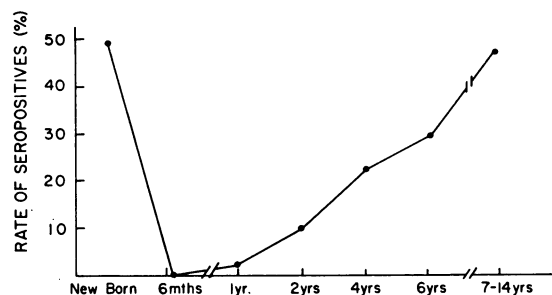


Fig. 1. Prevalence of *Toxoplasma* antibodies in 550 newborn infants, and in 330 children distributed in groups of 50 from 6 months to 14 years of age.

(Fig. 2) and that among the children over 2 years of age, high antibody levels were present in 2% of 6-year-old and in 14% of 7–14-year-old children (Table 2).

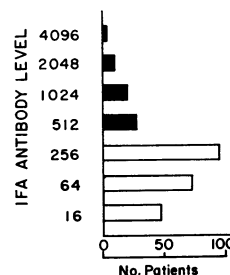


Fig. 2. Distribution of *Toxoplasma* antibody levels (i.e., reciprocals of serum dilution) in 550 newborn infants. (Unshaded are positive titres of low level; shaded are titres at high levels).

Table 2. Distribution of 250 children from different age groups by IFA antibody levels (i.e., reciprocal of serum dilution)

IFA anti-body level	Age groups (years)				
	1	2	4	6	7-14
<i>high</i>					
4 096	—	—	—	—	1
2 048	—	1	—	—	—
1 024	—	—	1	1	3
512	—	1	2	—	3
<i>low</i>					
256	—	—	3	4	3
64	—	—	2	4	6
16	1	3	3	7	8
no. positive	1	5	11	16	24
no. negative	49	45	39	34	26
total	50	50	50	50	50

The rate of seroconversion as a result of *Toxoplasma* infection in the first 2½ years of life was studied in 50 infants who at 6 months of age showed negative antibody results. A year later, 3 of 27 (11.1%) seroconverted, 1 with a titre of 1/16 384, and 2 with titres of 1/64. After 2 years of follow up, 5 of 35 (14.3%) were found to be positive, the 2 new cases having serologic titres of 1/256 and 1/64.

None of the patients with initially high serologic titres nor the ones that showed seroconversion had any of the characteristic features of clinical toxoplasmosis. The presence of enlarged lymph nodes was not different from that in seronegative patients and the adenopathy was never prominent enough to be called to our attention. In no instance was hepatosplenomegaly, a rash, or chorioretinitis observed.

#### Epidemiologic factors

Of the factors studied that could lead to infection with *Toxoplasma*, the results suggest a significant association with eating meat. Thus, 2 of the 5 infants that showed seroconversion ate raw or undercooked meat regularly as against only 1 of 30 seronegative children ( $p < 0.001$ ). Also, while 54% of the children were directly exposed to cats, no significant relationship could be established between this and the occurrence of seroconversion. The infrequency of garbage re-

moval and especially the presence of houseflies and cockroaches were common to the entire population studied, but again no direct association with the acquisition of infection could be established.

#### DISCUSSION

The incidence of *Toxoplasma* infection among infants and children in the present study is surprisingly high, beginning during the second year of life and increasing steadily through infancy and childhood to reach a plateau in adolescence. For the group of 35 children followed up for 2 years, a rate of 1 seroconversion per 168 persons per month of observation was obtained, which is strikingly different from reports from the USA. Warren & Dingle (22) investigated children from middle and upper class populations and reported only 5% seroconversion after 10 years of follow-up. Lamb & Feldman (11), in a low to lower middle class population, showed a 3% rate of conversion during the first 4 years of life with only 1 antibody conversion per 2 392 persons per month of observation.

They suggested that the highest risk for *Toxoplasma* infection occurred during adolescence since they obtained positive reactions in approximately 30% of subjects in their third decade of life. Our results are much closer to those reported by Paul (13) and Desmonts (2) on European infants in whom infection started in the second year of life and increased thereafter, or to the results that Remington (16) obtained in Central America with a 23% rate of infection for the first 5 years of life.

The similar proportions of positive sera, 48% and 49%, among those aged 7-14 years and in newborn children, the latter having acquired maternal antibodies, suggest that infection with *Toxoplasma* in this population decreases significantly after adolescence.

None of the children with high antibody levels nor the 5 who showed seroconversion had any symptoms or signs that could be attributed to clinical toxoplasmosis. This finding is not surprising as it was observed by others, including Feldman (5) and Warren & Dingle (22), and it suggests that toxoplasmosis is a rather uncommon clinical entity even in areas of the world where the rate of infection is high and seroconversion among young children is common.

Although no definite conclusion can be drawn from a small number of patients, our results suggest that eating raw or undercooked meat is probably

significantly associated with early infection, especially since in this population refrigerators were seldom available and meat could not be stored chilled or frozen. The risk of infection is high if encysted toxoplasmas are present in meat, as was observed by Rawal (15) and by Jacobs et al. (10) in the tissues of different animals, particularly in sheep and pigs.

The recent discovery of resistant oocysts of toxoplasma in the faeces of cats (7, 8, 17) would make faecal transmission from the cat a possible route of infection in man as well as in other animals. Recently, Wallace (20) reported that 20% of 1 023 cats in Hawaii had antitoxoplasma antibodies and that toxoplasma oocysts were recovered from the faeces of 0.5% of the cats, providing the first epidemiological evidence for possible transmission of infection in nature. Additional support for this hypothesis was that the proportions of positive sera differed significantly between subjects who had possessed a cat sometime during their life and those who had not (14). Cats were commonly present in the households of the children in our study but no direct relationship could be established with regard to their presence and the occurrence of seroconversion. Moreover, sandboxes in which the cats could deposit their faeces were seldom provided. No attempt was made to determine the rate of infection in cats or to identify the presence of toxoplasma oocysts in their faeces. Consequently, it has not been established

whether soil contamination should be considered a factor at risk.

The role of houseflies and cockroaches, which are common vectors for many human intestinal parasites, in the dissemination of toxoplasma oocysts from the faeces of cats was suggested by Wallace (19, 21). In experimental conditions, it was shown that flies and cockroaches carried the infective oocyst for at least 24 hours after being exposed to the infected faeces of cats, and the transmission of oocysts to mice was demonstrated. These vectors could therefore be directly involved in the transmission of *Toxoplasma* to man or other animals in populations where cats are common house pets and where flies and cockroaches present a real sanitary problem. If this proves to be the case, the infection could be acquired by the consumption of contaminated food or water and not necessarily as a result of close contact with cats.

In conclusion, this study provides evidence that in a population of a low socioeconomic level, *Toxoplasma* infection appeared during the second year of life and increased steadily thereafter to reach a plateau in adolescence. Eating undercooked meat seems to be associated with early infection, but no difference could be established with regard to exposure to cats. The role of vectors such as houseflies and cockroaches in the transmission of infection requires further investigation.

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## RÉSUMÉ

### INFECTION PAR LES TOXOPLASMES CHEZ LES ENFANTS DANS UN PAYS EN VOIE DE DÉVELOPPEMENT

On a étudié la prévalence de l'infection par *Toxoplasma gondii* chez 850 enfants en bonne santé appartenant à une collectivité de faible niveau socio-économique dans différents secteurs urbains de Santiago (Chili). On comptait parmi eux 550 nouveau-nés et 300 enfants qui ont été répartis en 6 groupes de 50 selon l'âge (6 mois, 1, 2, 4, 6 et 7-14 ans). Les anticorps anti-toxoplasmes ont été recherchés par la méthode colorimétrique de Sabin-Feldman et par l'épreuve d'immunofluorescence indirecte dont les résultats se sont révélés concordants dans 98% des cas.

On a décelé des anticorps dans 49% des échantillons de sérum à la naissance, 0% à 6 mois, 2% à 1 an, 10% à 2 ans, 22% à 4 ans, 32% à 6 ans et 48% dans le groupe d'âge 7-14 ans. Pour déterminer le rythme d'infection et le rôle de certains facteurs épidémiologiques, on a suivi 50 enfants, âgés à l'origine de 6 mois, pendant 2 ans. Le taux de séroconversion a atteint 11 et 14% respectivement après 1 et 2 ans, soit 1 séroconversion par 168 sujets et par mois d'observation. Aucun signe de toxoplasmose clinique n'a été relevé parmi les enfants initialement porteurs de titres

élevés d'anticorps ni parmi ceux qui ont présenté une séroconversion.

D'après les auteurs, il semble exister un lien direct entre l'infection précoce par le toxoplasme et la consommation de viande crue ou insuffisamment cuite. Par contre, le rôle

dans la transmission des chats et de vecteurs éventuels comme les mouches et les blattes n'a pu être établi et de nouvelles recherches sur ces aspects épidémiologiques sont nécessaires.

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